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## Turfgrass Response to Levels of Arsenate and Phosphate in the Soil

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TURFGRASS RESPONSE TO LEVELS OF ARSENATE  
AND PHOSPHATE IN THE SOIL

BY  
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Approved:

Charles R. Smith

A THESIS SUBMITTED IN PARTIAL FULFILLMENT OF THE  
REQUIREMENTS FOR THE DEGREE OF MASTER  
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IN  
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Dean of the Graduate School

UNIVERSITY OF RHODE ISLAND

1963

MASTER OF SCIENCE THESIS

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1963

## ABSTRACT

This thesis covers field, greenhouse and laboratory studies that were conducted to determine the effect of various soil phosphorus levels on the herbicidal properties of calcium and copper arsenates. These studies were conducted during 1961 and 1962.

The greenhouse study involved work with annual bluegrass (Poa annua L.) and crabgrass (Digitaria ischaemum Schreb.) grown on two soil types. Levels of soil phosphorus were adjusted to low, medium and high for each soil type and copper and calcium arsenates each at two rates were added to each soil. It was found that the low rate of calcium arsenate was as effective in controlling crabgrass and annual bluegrass as were the other treatments. Soil types and phosphorus levels did not affect the degree of arsenic injury.

The purpose of the field test was to determine the response of annual bluegrass (Poa annua L.), colonial bentgrass (Agrostis tenuis Sibth.), red fescue (Festuca rubra L.), Kentucky bluegrass (Poa pratensis L.) and smooth crabgrass (Digitaria ischaemum Schreb.) to a single conventional rate of calcium arsenate under two levels of soil phosphorus. Each plot was (a) seeded and treated with arsenic the same day, (b) seeded three weeks after arsenic treatment and (c) seeded six weeks after the arsenic application. The high phosphorus level generally promoted better cover and growth than the low phosphorus level but did not effect arsenic activity in any measured way. Arsenic significantly reduced germination, rate of establishment and growth in height.

Chemical analyses of plant tops and roots showed that soil phosphorus level did not affect uptake of arsenate or movement of arsenate in the soil.

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## I. INTRODUCTION

Turfgrass culture has been greatly improved through an understanding of the ecology of various associations in mixtures of species. Turfgrass is fertilized, artificially watered, cut intensively and treated with pesticides in order to develop a durable and attractive area. Crabgrass (Digitaria ischaemum Schreb.) and annual bluegrass (Poa annua L.) have been troublesome natural invaders on turfed areas in temperate regions for many years. To enable the desired grasses to favorably compete with such weeds, many selective chemical herbicides have been used.

Arsenic has been used on turfgrass as a chemical weed control with relatively good success in recent years. The most common source of arsenic for this purpose is calcium arsenate, used as a pre-emergence killer for crabgrass and annual bluegrass. The response of these weeds to this chemical has been unpredictable apparently for reasons of variable physical and chemical soil conditions. On some soils, the arsenic treatment is adequate to control or injure the weed but in other soils does not give satisfactory control. Researchers have investigated the reasons for these unpredictable reactions but have not completely resolved this problem. Since arsenic is relatively plentiful, an inexpensive element to produce and, frequently, shows great promise as an herbicide, it seems appropriate to continue studies with it.

The objective of this experiment was to investigate further the herbicidal properties of arsenic, using copper arsenate and calcium

arsenate as source materials. If positive predictions and recommendations could be given for the use of arsenic on turfgrass, a commendable contribution would be made to turfgrass culture.

## II. REVIEW OF LITERATURE

The literature on the use of arsenic as a fertilizer for turfgrass is limited. The only reference found in the literature is a report by the U.S. Department of Agriculture, Bureau of Plant Industry, in 1911. This report states that arsenic is a valuable element for the growth of turfgrass, and that it is necessary to apply it in the form of arsenic acid or arsenic trioxide. The report also states that the application of arsenic to turfgrass should be made in the form of a solution, and that the solution should be applied to the surface of the turfgrass. The report also states that the application of arsenic to turfgrass should be made in the form of a solution, and that the solution should be applied to the surface of the turfgrass.

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## II. REVIEW OF LITERATURE

Much research has been conducted in recent years in attempts to determine specifically the action of arsenic in plants and soils. Arsenic is a cheap and plentiful element and, in certain compounds, it has shown remarkable promise for herbicidal and insecticidal uses. The performance of the arsenical compounds is erratic at times with no precise reason. If arsenic, in its many and varied forms, could be used with predictability as an herbicide it could be a valuable tool in crop production and vegetation control.

In reviewing the arsenical research, one factor which appears many times overlooked but that actually ties in closely with arsenic and the soil-plant relationship is water. It is the prime solvent for most inorganic compounds and has been shown to influence arsenic solubility. Smith (20), in working with arsenical burn resulting from calcium arsenate on cotton, found that dew collected from cotton plants was alkaline in reaction. The solids found in the dew were chiefly carbonates of calcium and magnesium. Calcium arsenate digested in samples of this dew and in distilled water showed 8.7 per cent of the arsenic acid dissolved by the dews compared with 0.08 per cent dissolved by distilled water. Patten and O'Mera (16) worked with calcium, magnesium and lead arsenates in an effort to determine the influence of carbon dioxide on solubility. The solubilities of calcium and magnesium arsenates were greatly increased after boiling 24 hours in water



saturated with carbon dioxide. The lead arsenate was less soluble in the saturated carbon-dioxide water than in free distilled water. Distilled water has a smaller solubility influence than does either tap water or carbon-dioxide saturated water according to Mogendorff (13). In performing greenhouse tests, it may be advantageous to know the source of water, even if this is not measured quantitatively. Results in the greenhouse may not compare favorably with those in the field for this reason.

Other factors beside water relationships have been found to be responsible for variable results with arsenic. Gile (9) reported that soil acidity was not connected in any important degree to arsenic activity. Everett (8) in contrast found that a change in soil pH from 5.3 to 5.9 depressed arsenate absorption.

Gile (9) also reported that variation in arsenic activity may be due to soil type. His work on some South Carolina soils showed that arsenic injury to plants occurred more frequently on sandy soils than on heavier soil types. It appeared to him that the increased colloid content of the heavier textured soils was responsible for the difference in the action of arsenic. After considerable experimentation, he reported that arsenic toxicity was reduced in direct proportion to the quantity of colloidal material present. Welton and Carroll (25) found that lead arsenate was effective on the light colored but not on the dark colored Madison County soil, the latter being high in organic matter and clay particles.

Hurd-Karrer (11) experimented with sodium arsenate on a clay loam and found that recovery growth of arsenic-injured plants growing in soils containing excess phosphate was less as the plant entered the jointing

stage although it had not been apparent at younger stages. After a nine-week period, the plants were harvested and found to have a less toxic appearance where phosphate was applied. This was shown by greater dry weights and unretarded stages of development. The averaged plant weights showed that 125 and 250 ppm of arsenic reduced yields to 54 and 20 per cent respectively. Phosphate application corresponding to 125 ppm of phosphorus increased yields to 73 and 41 per cent over control. In another experiment, plants were grown in pots for a five-week period in a very sandy soil. The arsenic in these soils proved to be so toxic that 50 ppm severely injured the plants within a few weeks except where phosphate was added. "One hundred ppm of phosphorus definitely improved their condition, while 200 ppm of phosphorus made them indistinguishable from the control." In summary Hurd-Karrer states, "Thus the toxicity of sodium arsenate and the extent to which phosphate can inhibit it depends on the soil type."

Steckel (21) states:

Arsenate behaves chemically in a manner quite similar to phosphate. The soil chemistry of arsenate and phosphate is quite similar but not identical. We know that some soils fix large amounts of phosphate and that fertilizer phosphate applications must be large in order to feed adequately the growing plant. A high phosphate fixing soil will be also a high arsenate fixing soil. Low phosphate fixing soils respond to small amounts of phosphate fertilizer, so the low phosphate fixing soil will be a low arsenate fixer.

Jackson (12) found that arsenate and phosphate are alike chemically since both develop the typical molybdate color complex. Gile (9) reports that iron was the chief constituent of the colloid effecting the toxicity of calcium arsenate as it was in the phosphorus colloid. The soil colloid reduces the toxicity of calcium arsenate by forming an insoluble ferric arsenate when any arsenate ion becomes available. In working



with aluminum of the colloid, he found it to have no effect on the toxicity of calcium arsenate. He also found phosphate to have little effect on the arsenate toxicity as such, since the colloid had a greater affinity to fix the arsenate than the phosphate. Welton and Carroll (25) found, in contrast, that by applying phosphorus one month before lead arsenate was applied for pre-emergence crabgrass control, a reduction in the effectiveness of the arsenate was displayed. More crabgrass plants developed on the phosphorus pretreated plots than on the non-pretreated plots.

Studies by Dean and Rubens (5) show another factor to influence arsenic toxicity. "Soils treated by techniques which are essentially a counterpart of the base exchange methods show a definite anion exchange capacity." They found also that anions are not adsorbed by soils to the same degree. There is about twice as much phosphate adsorbed, as exchangeable anions, as there is arsenate. Arsenates will not displace all of the phosphates adsorbed by the soils, whereas the phosphates may displace most of the adsorbed arsenates. They reported that:

When soils are partially saturated with phosphorus under natural conditions by the long continued use of phosphate fertilizer in the field, apparently the exchangeable phosphorus is for the most part as readily replaceable with arsenate as with small anions such as flouride, whereas when soils are fully saturated under rather drastic laboratory conditions, only about half of the adsorbed phosphate is replaced by arsenate.

Swenson, Cole, and Sieling (22) observed that arsenates were found less capable than certain of the organic acids to replace phosphate on soil colloids.

Under conditions of identical concentrations of arsenate and phosphate, the phosphate was about five times as effective in replacing arsenate as was the arsenate in replacing phosphate from the chemically combined form. Those organic acids which were more effective in replacing phosphate than was arsenate, would then be very effective in replacing arsenate.

They also pointed out that since arsenates are displaced more readily than phosphates, arsenates would be displaced by phosphates and certain organic acids to lower horizons where the arsenate ion would be less detrimental to the shallow rooted crops. Hurd-Karrer (11) stated that in some South Carolina soils the phosphorus actually enhanced injury caused by calcium arsenate. This occurred by combining with the iron in the soil, making it unavailable for insoluble combination with the arsenic. Everett (8) in his investigation with bluegrass, found that phosphorus actually caused a small consistent increase in the plant arsenic. When the arsenate was applied to the bluegrass at 285 pounds per acre, yields were reduced 20 to 30 per cent in May but caused no visible injury during the remainder of the growing season.

Cations as well as anions have been found by Thomas (23) to effect the activity of arsenic in soils. He treated the clay minerals bentonite, illite, and kaolinite with arsenic and found that an increase in cation exchange capacity of the kaolinite took place by 54 per cent whereas cation exchange for bentonite and illite was slightly decreased. Other factors caused a penetration of arsenic to a greater depth than could be accounted for by mechanical mixing. "The movement of arsenate in the soil was related to the degree of base saturation and nature of cation. Larger amounts of arsenic were found at greater depths in soils having a high base saturation." Arsenic penetration increased in the order of H-Ca-Na after the soil was treated.

Although a knowledge of the soil and its selective influence is known for arsenic, this would be useless without accompanying information concerning the plants response to arsenic in the soil. Calcium arsenate is used presently as a pre-emergent weed control chemical.



Some have felt that arsenic is most effective as an inhibitor in seed germination. Welton and Carroll (25) after considerable experimentation with arsenicals as pre-emergence herbicides made the following assumption; in order for lead arsenate to control crabgrass it must be in contact with the seed for a considerable period of time during which it gradually breaks down, penetrates the seed coat and eventually kills the embryo of the seed. DeFrance (6) at the Rhode Island Experiment Station took a different approach in controlling crabgrass with arsenic. He considered the logical time to treat with chemicals to be during the period of seed head formation. When sodium arsenite was sprayed on the stand of crabgrass at seeding, a decrease in seed germination resulted.

The action of arsenic in the plant's processes is very similar to that of phosphorus. Daniel (3) reported that a low phosphorus availability level is required if arsenic is to inhibit Poa annua growth. Goetze (10) reported that, in soil containing low amounts of phosphorus and 12 to 16 pounds of calcium arsenate per 1000 square feet, the establishment of new Poa annua seedlings was prevented. He found that additional quantities of arsenic may be required to build up toxic concentrations where soils are high in phosphorus. In another article, Daniel (4) states that arsenic, although absorbed in a pattern similar to phosphorus, is not transferred from the area of absorption like phosphorus. Arsenic seems to be relatively immobile and, therefore, at times would cause a phosphorus deficiency symptom. He also observed that "young plant roots take up arsenic and combine it to the carbohydrate metabolism of the plant, replacing some of the phosphorus normally present in the carbohydrate molecules." Ahlgren, Klingman, and Wolf (1) state that proteins in the plant protoplasm are denatured or



precipitated by arsenic ions. Rogers (17) reported that arsenate chemically and physically resembles the phosphate group in plant metabolism by interrupting the conservation and transfer of bond energies. "For instance, glucose-1-phosphate along with sucrose phosphorylase and fructose in a solution can produce sucrose. If arsenate is present, glucose-1-arsenate is produced and spontaneously hydrolyzes to glucose and arsenate and no sucrose is found."

Other workers have resorted to nutrient cultures to answer certain arsenic-phosphorus relationships. Everett (8) states:

High phosphorus in the solution eliminated visible arsenate injury to bluegrass and crabgrass. Also, an addition of phosphorus to 100 ppm reduced arsenate absorption by crabgrass from 246 to 29 ppm of arsenic. The absorption of arsenate by bluegrass was reduced more by 10 ppm than by 1 or 100 ppm of phosphorus in the solution.

Hurd-Karrer (11) came to the conclusion, after working with Hard Federation wheat in varying phosphate-arsenate nutrient solutions, that a 1:1 arsenic:phosphorus ratio was almost lethal, a 1:2 ratio highly toxic, while a 1:5 ratio rendered the arsenic harmless. Rumburg, Engel and Meggitt (18) worked with Clinton oats and found that when phosphorus levels of 10 and 62 ppm were maintained in nutrient solutions, the uptake of arsenate was inhibited with no arsenic poisoning being observed. They also found that 10 ppm of arsenic in a solution of 1 ppm of phosphorus caused the oats to become flacid 24 to 48 hours after addition of the arsenate and were near death after 4 days. They state that "The uptake of arsenic after 72 hours at 1 ppm phosphorus was 1.9 times greater than at 10 ppm phosphorus and 7.5 times greater than 62 ppm phosphorus."

In summary, there still seems to be much unknown about the mechanism of arsenic activity in soils as well as in plant nutrition. Hurd-Karrer (11) states, "It is thus probable that the varying condition for solubility, adsorption, and chemical combination of arsenic and phosphorus in soils render unpredictable the practical value of the arsenic phosphorus antagonism." An attempt will be made in this thesis to contribute further knowledge to this yet unanswered question of arsenic activity as a pre-emergent chemical.

### III. THE INVESTIGATION

#### A. Objective

The major objective was to determine whether various levels of soil phosphorus would affect the herbicidal properties of calcium and copper arsenates. The investigation included a greenhouse study, a field study and chemical determinations on the plant material obtained from the field study. The chemical determinations included a test for total arsenic and total phosphorus in an effort to observe any difference in element assimilation. Soil tests were also made on the field test soils to observe the movement of arsenic in the soil under two levels of phosphorus.

#### B. Greenhouse Pot Test

1. Purpose. The purpose of this test was to determine the effects of copper arsenate and calcium arsenate on crabgrass (Digitaria ischaemum Schreb.) and annual bluegrass (Poa annua L.) grown on two soil types with three levels of phosphorus. In previous studies by DeFrance and Kollett (7) at Rhode Island, copper compounds showed considerable promise for the control of annual bluegrass. Since both copper and arsenic have indicated some control of both annual bluegrass and crabgrass, it seemed feasible to investigate compounds containing both elements. The literature review has pointed out differential grass response to arsenic based on soil type and soil phosphorus content. It was hoped that



through the greenhouse study, certain answers could be obtained that would lead to a more precise field test.

2. Materials and Methods. The experiment was established using four-inch plastic pots arranged in three blocks, with each block representing one replication. Included in the blocks were the two grasses planted in each of two soils. The soils were Bridgehampton silt loam and a fine sandy loam. Each soil was then partitioned into three segments with each segment representing a low, medium and high phosphorus level. Calcium arsenate and copper arsenate, each at two rates, were applied on both grasses.

Viable seed of both grasses was obtained and the per cent germination was established. One hundred seeds of each grass were counted for planting in each pot.

To bring the soil partitions to a medium and high level of phosphorus, superphosphate was added and mixed by hand. The final levels of available phosphorus were:

1. silt - high phosphorus	135 ppm
2. silt - medium phosphorus	82 ppm
3. silt - low phosphorus	19 ppm
4. sand - high phosphorus	137 ppm
5. sand - medium phosphorus	87 ppm
6. sand - low phosphorus	13 ppm

These levels were designated by the well-known Truog method for readily available phosphorus (24). Since the soil was low in pH, sufficient hydrated lime was added to bring final pH to about 6.1 for both soils. Lime was added 12 days before the phosphorus to avoid fixation of

phosphorus by calcium ions. Copper arsenate was applied to the soil surface at rates of 888 and 444 pounds per acre. This material analyzed 91 per cent active copper arsenate and contained not less than 24.2 per cent total arsenic. The calcium arsenate was applied at 783 and 391 pounds per acre. This arsenical was guaranteed to have 73 per cent active calcium arsenate by weight and to contain not less than 27.5 per cent total arsenic. The pots were then placed in a 35°F. cool room as suggested by Welton and Carroll (25) who indicated that crabgrass injury occurred only after arsenic had had ample time to penetrate the seed. While in the cool room, the pots were rotated systematically to remove any possible variation caused by persistent differing micro-climates. The pots were not allowed to become dry at any time during this period. Two months after the pots had been placed in the cool room, they were removed and placed at random in the greenhouse. Again the pots were kept moist during germination and throughout the remainder of the test. Germination percentage and height-of-growth data were obtained for the two grass species.

3. Results and Discussion. Prior research at this station (7) (2) has shown a definite effect on both annual bluegrass and crabgrass following soil applications of calcium arsenate and certain copper compounds. Although copper arsenate as a single compound has not reportedly been used before to control annual bluegrass and crabgrass as such, previous literature shows the two components of the compound to have an herbicidal controlling potential.

An effort was made to look at each of the grasses as single variances and they will be discussed as such. The first factor observed was germination. Four days after the pots were placed in the greenhouse,



some crabgrass germination was observed. Final germination readings were taken nine days later. The results of germination data will not be given, however, since the grasses averaged between 85 and 100 per cent germination with no consistent obvious differences between treatments. All grass in the arsenate treated soil showed phosphorus deficiency. A purple leaf coloration, usually associated with phosphorus deficiency, was present for the duration of the study. Daniel (4) reported that high arsenate causes phosphorus deficiency because the arsenate replaces some of the normally adsorbed phosphate but does not satisfy the plant's requirement for phosphate.

One month after moving the pots to the greenhouse, the height of annual bluegrass was recorded in each pot and this data was analyzed and is reported in Table I. The height in growth for the check was significantly taller than any of the other treatments. Calcium arsenate at 783 pounds per acre gave a significant reduction in growth over the other treatments. With respect to soils, the silt loam was significantly better in supporting growth than was the sandy loam. The grass height on the high phosphorus-level soils was significantly greater than that on the low and medium levels. There was no interaction between treatments. The high phosphorus level did not overcome the arsenate injury as might be expected from the literature, as compared with the check plots.

The analysis of variance on the height of crabgrass showed results similar to those for annual bluegrass with the exception of one significant interaction. The crabgrass growth response is given in Table II. The check plot, as in the previous table, showed significant

TABLE I

AVERAGE HEIGHT IN CENTIMETERS OF ANNUAL BLUEGRASS  
30 DAYS AFTER GERMINATION ON ARSENATE TREATED  
SOILS WITH THREE PHOSPHORUS LEVELS

	Sandy Loam			Silt Loam			Average			
Chemical Treatment	Phos. Levels			Phos. Levels			Phos. Levels			Treat. Av.
	L	M	H	L	M	H	L	M	H	
$\text{Cu}_3(\text{AsO}_4)_2$ 888 lb./A	0.7	1.0	0.9	2.0	3.2	3.5	1.3	2.1	2.2	1.9
$\text{Cu}_3(\text{AsO}_4)_2$ 444 lb./A	1.6	3.0	4.3	2.3	2.5	4.0	1.9	2.8	4.2	3.0
$\text{Ca}_3(\text{AsO}_4)_2$ 783 lb./A	0	0	0	0.5	0.5	0	0.2	0.2	0	0.2
$\text{Ca}_3(\text{AsO}_4)_2$ 391 lb./A	2.7	2.2	1.0	1.5	1.3	3.5	2.1	1.8	2.2	2.0
Check	4.9	5.6	7.2	5.2	6.3	7.0	5.0	6.0	7.1	6.0
Average	<u>1.9</u>	<u>2.4</u>	<u>2.7</u>	<u>2.3</u>	<u>2.8</u>	<u>3.6</u>	<u>2.1</u>	<u>2.6</u>	<u>3.1</u>	2.6
Soil Type Av.	2.3			2.9						

## Summary of Analysis of Variance

Source of Variation	F Value	D Value 5%
Chemical Treatment	48.9	1.2
Soil Type	4.0	0.6
Phosphorus Level	4.6	0.9
Treatment x Soil	1.9	N.S.
Treatment x Phosphorus	1.1	N.S.
Phosphorus x Soil	0.5	N.S.
Treatment x Soil x Phosphorus	1.0	N.S.
Coefficient of Variability - 49.98%		

TABLE II

AVERAGE HEIGHT IN CENTIMETERS OF CRABGRASS 30 DAYS  
AFTER GERMINATION ON ARSENATE TREATED SOILS  
WITH THREE PHOSPHORUS LEVELS

	Sandy Loam			Silt Loam			Average			
Chemical Treatment	Phos. Levels			Phos. Levels			Phos. Levels			Treat. Av.
	L	M	H	L	M	H	L	M	H	
$\text{Cu}_3(\text{AsO}_4)_2$ 888 lb./A	0.7	1.5	1.5	0.6	0.4	0.7	0.6	0.9	1.1	0.9
$\text{Cu}_3(\text{AsO}_4)_2$ 444 lb./A	0.3	0.8	0.5	0.9	0.8	0.8	0.6	0.8	0.7	0.7
$\text{Ca}_3(\text{AsO}_4)_2$ 783 lb./A	0.7	1.7	1.8	0	0	0	0.3	0.8	0.9	0.7
$\text{Ca}_3(\text{AsO}_4)_2$ 391 lb./A	0.1	0	0	0	0.4	0.6	0.1	0.2	0.3	0.2
Check	5.8	7.3	8.2	6.0	7.2	8.1	5.9	7.2	8.1	7.1
Average	<u>1.5</u>	<u>2.3</u>	<u>2.4</u>	<u>1.5</u>	<u>1.8</u>	<u>2.0</u>	<u>1.5</u>	<u>2.0</u>	<u>2.2</u>	1.9
Soil Type Av.	2.1			1.8						

## Summary of Analysis of Variance

Source of Variation	F Value	D Value 5%
Chemical Treatment	251.4	0.3
Soil Type	3.2	N.S.
Phosphorus Level	6.6	1.0
Treatment x Soil	4.0	1.0
Treatment x Phosphorus	1.9	N.S.
Phosphorus x Soil	0.7	N.S.
Treatment x Soil x Phosphorus	0.4	N.S.
Coefficient of Variability - 12.81%		



growth of crabgrass compared with the other treatments. The other treatments reacted in a similar manner with no significance between treatments. No significant differences were found between soil types for the support of growth. The high and medium phosphorus level was significant in increasing plant growth over the low level; however, no significance between high and medium phosphorus levels was evident.

To investigate the significant interaction of treatment x soil, the following table of averages is given.

TABLE III

## INTERACTION OF TREATMENT X SOIL

	$\text{Cu}_3(\text{AsO}_4)_2$ 888 lb./A	$\text{Cu}_3(\text{AsO}_4)_2$ 444 lb./A	$\text{Ca}_3(\text{AsO}_4)_2$ 783 lb./A	$\text{Ca}_3(\text{AsO}_4)_2$ 391 lb./A	Check
Sandy loam	1.2	0.7	1.4	0.11	7.1
Silt loam	0.6	0.9	0	0.31	7.1
					F value = 4.0
					D value 5% = 1.0

Calcium arsenate at 783 pounds per acre was the only interaction obviously different. There was a definite decrease in height due to silt loam under the 783 pound per acre rate.

The results of this experiment gave insight into certain factors that could be eliminated in the field test. Calcium arsenate at the 391 pound-per-acre rate appeared to be sufficiently effective in reducing weed growth over the copper compound. The silt loam supported better or equally as good growth as did the sandy loam with the phos-

phorus being different only at low and high rates or levels.

### C. Field Plot Experiment

1. Purpose. In the spring of 1962, an investigation similar to the greenhouse test was established on the University of Rhode Island Turf Plots. After analyzing the results of the greenhouse test, it was determined that a different approach might be taken to answer the major objective. The specific purpose of this experiment was to observe the response of five different grasses to a single rate of calcium arsenate under two levels of soil phosphorus. An effort was made to set up variables which would resemble the procedures most often used on lawn turf. Since varying recommendations are made relative to time of application for calcium arsenate, it was decided that three different seeding dates would be employed following the initial application of calcium arsenate.

2. Materials and Methods. The soil on which the experiment was established was a Bridgehampton silt loam. The test area soil had an available phosphorus level of 12.4 ppm by the Truog method (24), which is considered very low. The source of calcium arsenate used was the same as that in the pot test. To keep within the range of conventional rates used for most turf grass applications, 391 pounds per acre of calcium arsenate was used. The five grasses chosen were the three most often used for turf establishment in the Northeast and the two weed grasses used in the greenhouse test. They included; annual bluegrass (Poa annua L.), Colonial bentgrass (Agrostis tenuis Sibth.), Red fescue (Festuca rubra L.), Kentucky bluegrass (Poa pratensis L.) and smooth



crabgrass (Digitaria ischaemum Schreb.). Superphosphate and dolomitic limestone were used for phosphorus and calcium sources respectively.

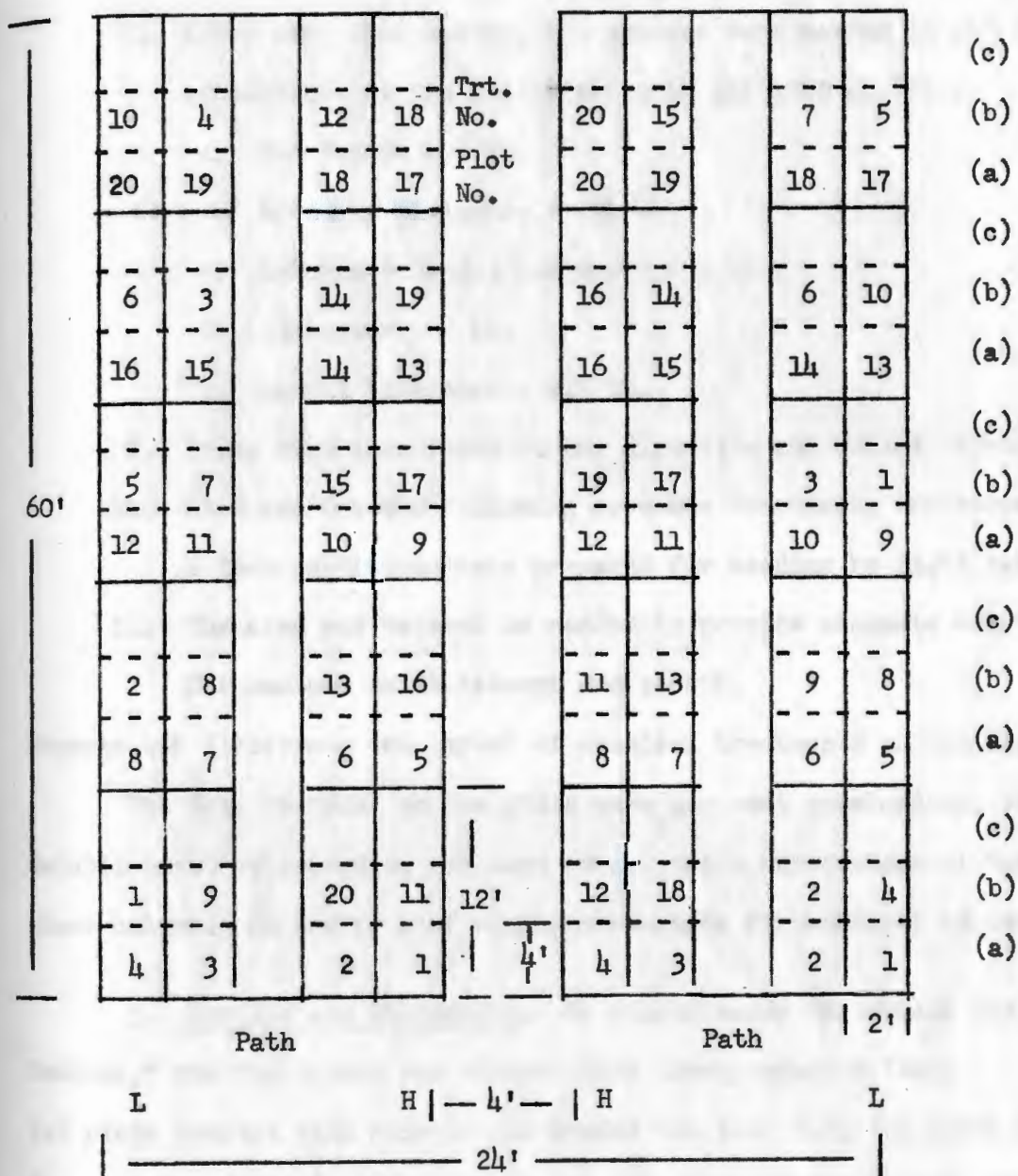
The plot layout was established by using four blocks, each representing a replication. The blocks were then split into low and high phosphorus strips, making them the whole plots. The subplots consisted of the five grasses, each having a calcium arsenate and no arsenate treatment. These were completely randomized within each of the whole plots. The individual plots were 2 x 12 divided into three 4-foot segments for the respective seeding dates. The respective layout is shown in detail in Figure 1.

The following establishment procedures were used. The area was prepared for seeding as recommended by the U.R.I. Agricultural Extension Service Bulletin No. 183, (19). In general the procedure was as follows:

1. The area was first rough graded by hand.
2. Limestone was added at a rate of 115 pounds per 1000 sq. ft. and tilled to a depth of 6 inches.
3. Muriate of potash (KCl) was added at the rate of 3.34 pounds per 1000 sq. ft., also 12.5 pounds of sodium nitrate as the nitrogen source.
4. Superphosphate was applied at a rate of 130 pounds per 1000 sq. ft. giving final phosphorus levels of 135 ppm for the high level and 20 ppm for the low.
5. These were all tilled to a depth of 6 inches one week after the limestone to avoid excess fixation by the calcium ions.
6. The final seedbed was prepared by rolling and raking, being careful not to move soil from one phosphate strip to another.

BLOCK II

BLOCK I



- (c) Seeded 6 weeks after treatment.  
 (b) Seeded 3 weeks after treatment.  
 (a) Seeded and treated same day.

FIGURE 1

TWO OF THE FOUR BLOCKS WITH NUMBER RANDOMIZATION  
 OF PLOTS AND TREATMENTS



7. Calcium arsenate was applied at 8.9 pounds of active arsenic per 1000 sq. ft. to the respective treatment plots.
8. After very fine raking, the grasses were seeded in one set of subplots at the following rates per 1000 sq. ft.:
  - a. Red fescue - 5 lb.
  - b. Kentucky bluegrass - 2 lb.
  - c. Astoria colonial bentgrass - 1 lb.
  - d. crabgrass - 2 lb.
  - e. annual bluegrass - 2.5 lb.
9. These were then raked in one direction and rolled lightly.
10. At 3 and 6 weeks following arsenate treatment, the respective 4-foot partitions were prepared for seeding by light raking.
11. The area was watered as needed to provide adequate moisture for maximum establishment and growth.

Figures 2-5 illustrate the layout of chemical treatments on the field plots.

The data recorded on the plots were per cent germination, rates of establishment by recording per cent cover, and a measurement of the plant height. An analysis of variance was made for each set of data.

3. Results and Discussion. As stated under "Materials and Methods," the field test was divided into three separate tests: (a) plots treated with arsenic and seeded the same day, (b) plots seeded 3 weeks after arsenic application, and (c) plots seeded 6 weeks after arsenic treatment. Only general differences and similarities will be reported among the seeding dates.

As shown in Table IV for test (a), germination was affected by the calcium arsenate application. There was no difference due to soil



FIGURE 2

GENERAL VIEW OF CHEMICAL TREATMENT



FIGURE 3

GENERAL LAYOUT OF BLOCK II





FIGURE 4  
CLOSE-UP VIEW OF ONE TREATMENT



FIGURE 5  
SIX-WEEK SEEDING READY FOR ESTABLISHMENT

TABLE IV

AVERAGE PER CENT GERMINATION OF GRASSES SEEDED AND  
TREATED SAME DAY WITH ARSENATE UNDER TWO  
PHOSPHORUS LEVELS

Grasses	High Phosphorus		Low Phosphorus		Treatment Average		Species Average
	Arsenic Treated	Check	Arsenic Treated	Check	Arsenic Treated	Check	
Annual Bluegrass	91.2	93.8	88.8	92.5	90.0	93.1	91.6
Colonial Bentgrass	77.5	82.5	75.0	90.0	76.2	86.2	81.2
Red Fescue	70.0	78.8	67.5	86.2	68.8	82.5	75.6
Kentucky Bluegrass	42.5	61.2	51.2	47.5	46.9	54.4	50.6
Crabgrass	80.0	100.0	87.5	100.0	83.8	100.0	91.9
Average	<u>72.2</u>	<u>83.2</u>	<u>74.0</u>	<u>83.2</u>	<u>73.1</u>	<u>83.2</u>	78.2
P Level Av.	77.8		78.6				

#### Summary of Analysis of Variance

Source of Variation	F Value	D Value 5%
Phosphorus Levels	0.1	N.S.
Chemical Treatment	19.1	4.6
Grass Species	42.6	10.3
Phosphorus x Chemical	0.1	N.S.
Phosphorus x Grass	0.3	N.S.
Chemical x Grass	1.0	N.S.
Phosphorus x Chemical x Grass	1.8	N.S.
Coefficient of Variability - 13.24%		

phosphorus level and no interaction between each respective source of variation. Each grass response was different, with annual bluegrass and crabgrass germinating significantly faster than the other three species. Fescue germinated significantly better than Kentucky bluegrass. In general it may be stated that germination of colonial bentgrass and Kentucky bluegrass appeared to be decreased the most, annual bluegrass and crabgrass the least, with red fescue falling intermediately.

The figures representing averages of the sources of variation on rate of establishment are shown in Table V. All major factors were significant at the 5 per cent level. The high soil phosphorus level promoted better growth; the arsenic treatment was significant in reducing rate of establishment over the control; and the grasses responded significantly different to both treatments. The significant interaction between phosphorus and grasses may be explained by the following table:

TABLE VI

INTERACTION OF PHOSPHORUS X GRASSES FOR RATE OF ESTABLISHMENT

	Low Phosphorus	High Phosphorus
Annual bluegrass	63.8	85.6
Colonial bentgrass	54.4	70.0
Red fescue	63.2	69.4
Kentucky bluegrass	40.0	52.5
Crabgrass	55.0	86.2
		F value = 3.4
		D value 5% = 10.3



TABLE V

AVERAGE PER CENT COVER OF GRASSES SEEDED AND  
TREATED SAME DAY WITH ARSENATE UNDER  
TWO PHOSPHORUS LEVELS

Grasses	High Phosphorus		Low Phosphorus		Treatment Average		Species Average
	Arsenic Treated	Check	Arsenic Treated	Check	Arsenic Treated	Check	
Annual Bluegrass	83.8	87.5	58.8	68.8	71.2	78.1	74.7
Colonial Bentgrass	67.5	72.5	47.5	61.2	57.5	66.9	62.2
Red Fescue	61.2	77.5	57.5	68.8	59.4	73.1	66.2
Kentucky Bluegrass	46.2	58.8	35.0	45.0	40.6	51.9	46.2
Crabgrass	80.0	92.5	45.0	65.0	62.5	78.8	70.6
Average	<u>67.8</u>	<u>77.8</u>	<u>48.8</u>	<u>61.8</u>	<u>58.2</u>	<u>69.8</u>	64.0
P Level Av.	72.8		55.2				

## Summary of Analysis of Variance

Source of Variation	F Value	D Value 5%
Phosphorus Levels	17.6	13.3
Chemical Treatment	24.8	4.6
Grass Species	18.0	10.2
Phosphorus x Chemical	0.4	N.S.
Phosphorus x Grass	3.4	10.3
Chemical x Grass	0.5	N.S.
Phosphorus x Chemical x Grass	0.3	N.S.
Coefficient of Variability - 16.16%		



Annual bluegrass and crabgrass showed a significant increase in rate of establishment under high phosphorus as compared with the remaining species.

In discussing the height measurements, as given in Table VII, the data are similar to those for rate of establishment. High phosphorus soil level was significant in enhancing growth over the control plots. There was also a significant difference in height among grass species. Crabgrass was significantly taller than the other grasses and annual bluegrass was taller than any of the basic turf grasses. The interaction of phosphorus x grasses is shown in Table VIII.

TABLE VIII  
INTERACTION OF PHOSPHORUS X GRASSES FOR HEIGHT

	Low Phosphorus	High Phosphorus
Annual bluegrass	5.6	10.0
Colonial bentgrass	4.5	8.2
Red fescue	4.8	6.0
Kentucky bluegrass	3.1	6.8
Crabgrass	6.3	12.5
		F value = 5.9
		D value 5% = 0.5

The high phosphorus plots showed a definite effect in increasing growth of all grass species, particularly annual bluegrass and crabgrass. As may be noted, this interaction was approximately the same as for the previous rate of establishment data.

TABLE VII

AVERAGE HEIGHT IN CENTIMETERS OF GRASSES SEEDED  
AND TREATED SAME DAY WITH ARSENATE  
UNDER TWO PHOSPHORUS LEVELS

Grasses	High Phosphorus		Low Phosphorus		Treatment Average		Species Average
	Arsenic Treated	Check	Arsenic Treated	Check	Arsenic Treated	Check	
Annual Bluegrass	9.2	10.8	5.0	6.2	7.1	8.5	7.8
Colonial Bentgrass	8.0	8.5	4.2	4.8	6.1	6.6	6.4
Red Fescue	5.2	6.8	4.0	5.8	4.6	6.2	5.4
Kentucky Bluegrass	5.2	8.5	2.8	3.5	4.0	6.0	5.0
Crabgrass	11.2	13.8	5.5	7.0	8.4	10.4	9.4
Average	<u>7.8</u>	<u>9.6</u>	<u>4.3</u>	<u>5.4</u>	<u>6.0</u>	<u>7.6</u>	6.8
P Level Av.	8.7		4.9				

## Summary of Analysis of Variance

Source of Variation	F Value	D Value 5%
Phosphorus Levels	12.4	3.5
Chemical Treatment	19.6	0.7
Grass Species	22.5	1.5
Phosphorus x Chemical	1.1	N.S.
Phosphorus x Grass	5.9	0.5
Chemical x Grass	0.7	N.S.
Phosphorus x Chemical x Grass	0.5	N.S.
Coefficient of Variability - 22.30%		

In summary, when the five grasses were treated with arsenic and seeded the same day, phosphorus had no effect on germination but did have an effect on rate of establishment and growth in height. The arsenic had an inhibiting effect on all three measurements. Each grass responded differently under each type of test; however, crabgrass and annual bluegrass showed the greatest differences in response to the treatments. The phosphorus levels reacted independently of the arsenic treatment in promoting a significantly greater rate of establishment and height of growth with no obvious interaction. Welton and Carroll (25) reported that arsenic is not readily soluble and needs a certain interval of time between application and germination to effectively control seed germination and growth.

Growth on series (b), or those plots seeded three weeks after arsenic application, appeared to be more erratic and variable. Averages for germination are given in Table IX. The only significant source of variation for germination was the grasses. Neither phosphorus level nor chemical treatment with arsenic effected the germination. Annual bluegrass and crabgrass germinated significantly better than did either colonial bentgrass and Kentucky bluegrass. Red fescue was intermediate, being better than Kentucky bluegrass.

The rate of establishment data in Table X shows that high phosphorus was not significant in promoting a denser cover. The arsenic treatment had no significant effect in reducing rate of establishment; however, the grasses were different with annual bluegrass showing the fastest establishment. Crabgrass establishment was inhibited less than either colonial bentgrass or Kentucky bluegrass.



TABLE IX

AVERAGE GERMINATION OF GRASSES SEEDED THREE WEEKS  
AFTER ARSENATE TREATMENT UNDER  
TWO PHOSPHORUS LEVELS

Grasses	High Phosphorus		Low Phosphorus		Treatment Average		Species Average
	Arsenic Treated	Check	Arsenic Treated	Check	Arsenic Treated	Check	
Annual Bluegrass	76.2	77.5	73.8	75.0	75.0	76.2	75.6
Colonial Bentgrass	65.0	67.5	58.8	61.2	61.9	64.4	63.1
Red Fescue	68.8	70.0	70.0	72.5	69.4	71.2	70.3
Kentucky Bluegrass	60.0	58.8	58.8	58.8	59.4	58.8	59.1
Crabgrass	72.5	82.5	67.5	71.2	70.0	76.9	73.4
Average	<u>68.5</u>	<u>71.2</u>	<u>65.8</u>	<u>67.8</u>			68.3
P Level Av.	69.9		66.8				

Summary of Analysis of Variance

Source of Variation	F Value	D Value 5%
Phosphorus Levels	2.9	N.S.
Chemical Treatment	1.7	N.S.
Grass Species	11.8	8.5
Phosphorus x Chemical	0.1	N.S.
Phosphorus x Grass	1.0	N.S.
Chemical x Grass	0.5	N.S.
Phosphorus x Chemical x Grass	0.2	N.S.
Coefficient of Variability - 11.92%		

TABLE X

AVERAGE PER CENT COVER OF GRASSES SEEDED THREE WEEKS  
AFTER ARSENATE TREATMENT UNDER  
TWO PHOSPHORUS LEVELS

Grasses	High Phosphorus		Low Phosphorus		Treatment Average		Species Average
	Arsenic Treated	Check	Arsenic Treated	Check	Arsenic Treated	Check	
Annual Bluegrass	81.2	76.2	57.5	70.0	69.4	73.1	71.2
Colonial Bentgrass	43.8	45.0	33.8	42.5	38.8	43.8	41.2
Red Fescue	52.5	51.2	45.0	48.8	48.8	50.0	49.4
Kentucky Bluegrass	33.8	37.5	37.5	27.5	35.6	32.5	34.1
Crabgrass	55.0	68.8	40.0	43.8	47.5	56.2	51.9
Average	<u>53.2</u>	<u>55.8</u>	<u>42.8</u>	<u>46.5</u>	<u>48.0</u>	<u>51.1</u>	49.6
P Level Av.	54.5		44.6				

#### Summary of Analysis of Variance

Source of Variation	F Value	D Value 5%
Phosphorus Levels	4.3	N.S.
Chemical Treatment	1.9	N.S.
Grass Species	30.0	10.2
Phosphorus x Chemical	0.1	N.S.
Phosphorus x Grass	2.0	N.S.
Chemical x Grass	0.7	N.S.
Phosphorus x Chemical x Grass	1.6	N.S.
Coefficient of Variability - 20.67%		

Growth in height for grasses is shown in Table XI. The high phosphorus level was again significant in producing better growth in height. Arsenic had no significant effect while the grass species showed their usual differences. Crabgrass was significantly taller than the other four grasses. Annual bluegrass and colonial bentgrass were significantly taller than red fescue and Kentucky bluegrass.

In summary, neither the arsenic treatment nor the phosphorus levels were significant factors for germination or rate of establishment. Phosphorus did promote better growth in height. The grasses responded similarly to both phosphorus and arsenic for all three measurements. There were no significant interactions between major factors.

The last test to be discussed is the six-week seeding. Germination, Table XII, was not effected by phosphorus level, while the arsenic treatment caused considerable difference in germination and growth of the grasses. The average of germination for all grasses is lower for this seeding than either treatment (a) or (b). The grasses responded differently with fescue appearing to respond the least to arsenate toxicity and the most to phosphorus. Annual bluegrass and crabgrass were alike in germination rates with colonial bentgrass and Kentucky bluegrass significantly less.

Arsenic had a significant effect on rate of establishment for the six-week seeding as it did for germination. The phosphorus had no significant influence in promoting rate of establishment. These averages are found on Table XIII. The usual grass differences were present with annual bluegrass being more tolerant of soil factors than the other grasses.

The average height in growth for this time interval is somewhat



TABLE XI

AVERAGE HEIGHT IN CENTIMETERS OF GRASSES SEEDED  
THREE WEEKS AFTER ARSENATE TREATMENT  
UNDER TWO PHOSPHORUS LEVELS

Grasses	High Phosphorus		Low Phosphorus		Treatment Average		Species Average
	Arsenic Treated	Check	Arsenic Treated	Check	Arsenic Treated	Check	
Annual Bluegrass	10.2	9.0	7.8	8.8	9.0	8.9	8.9
Colonial Bentgrass	10.0	9.2	6.0	6.8	8.0	8.0	8.0
Red Fescue	8.0	5.5	7.2	7.0	7.6	6.2	6.9
Kentucky Bluegrass	4.8	5.8	3.8	3.5	4.2	4.6	4.4
Crabgrass	19.5	16.0	13.2	14.2	16.4	15.1	15.8
Average	<u>10.5</u>	<u>9.1</u>	<u>7.6</u>	<u>8.0</u>	<u>9.0</u>	<u>8.6</u>	8.8
P Level Av.	9.8		7.8				

#### Summary of Analysis of Variance

Source of Variation	F Value	D Value 5%
Phosphorus Levels	25.9	1.2
Chemical Treatment	0.5	N.S.
Grass Species	32.8	2.9
Phosphorus x Chemical	1.9	N.S.
Phosphorus x Grass	1.3	N.S.
Chemical x Grass	0.2	N.S.
Phosphorus x Chemical x Grass	0.4	N.S.
Coefficient of Variability - 30.94%		

TABLE XII  
AVERAGE GERMINATION OF GRASSES SEEDED SIX WEEKS  
AFTER ARSENATE TREATMENT UNDER  
TWO PHOSPHORUS LEVELS

Grasses	High Phosphorus		Low Phosphorus		Treatment Average		Species Average
	Arsenic Treated	Check	Arsenic Treated	Check	Arsenic Treated	Check	
Annual Bluegrass	55.0	75.0	61.2	72.5	58.1	73.8	65.9
Colonial Bentgrass	51.2	45.0	40.0	58.8	45.6	51.9	48.8
Red Fescue	70.0	80.0	67.5	80.0	68.8	80.0	74.4
Kentucky Bluegrass	42.5	36.2	33.8	46.2	38.1	41.2	39.7
Crabgrass	42.5	83.8	60.0	82.5	51.2	83.1	67.2
Average	<u>52.2</u>	<u>64.0</u>	<u>52.5</u>	<u>68.0</u>	<u>52.4</u>	<u>66.0</u>	59.2
P Level Av.	58		60.2				

Summary of Analysis of Variance

Source of Variation	F Value	D Value 5%
Phosphorus Levels	0.3	N.S.
Chemical Treatment	11.8	8.0
Grass Species	10.5	17.7
Phosphorus x Chemical	0.2	N.S.
Phosphorus x Grass	0.2	N.S.
Chemical x Grass	1.6	N.S.
Phosphorus x Chemical x Grass	1.1	N.S.
Coefficient of Variability - 3.00%		

TABLE XIII

AVERAGE PER CENT COVER FOR GRASSES SEEDED SIX WEEKS  
AFTER ARSENATE TREATMENT UNDER  
TWO PHOSPHORUS LEVELS

Grasses	High Phosphorus		Low Phosphorus		Treatment Average		Species Average
	Arsenic Treated	Check	Arsenic Treated	Check	Arsenic Treated	Check	
Annual Bluegrass	38.8	67.5	53.8	65.0	46.2	66.2	56.2
Colonial Bentgrass	43.8	51.2	30.0	50.0	36.9	50.6	43.8
Red Fescue	46.2	56.2	41.2	45.0	43.8	50.6	47.2
Kentucky Bluegrass	33.8	40.0	23.8	42.0	28.8	41.0	34.9
Crabgrass	18.8	53.8	25.0	40.0	21.9	46.9	34.4
Average	<u>36.2</u>	<u>53.8</u>	<u>34.8</u>	<u>48.4</u>	<u>35.5</u>	<u>51.1</u>	43.3
P Level Av.	45.0		41.6				

## Summary of Analysis of Variance

Source of Variation	F Value	D Value 5%
Phosphorus Levels	0.8	N.S.
Chemical Treatment	25.1	6.2
Grass Species	6.8	13.9
Phosphorus x Chemical	0.4	N.S.
Phosphorus x Grass	0.7	N.S.
Chemical x Grass	1.0	N.S.
Phosphorus x Chemical x Grass	1.3	N.S.
Coefficient of Variability - 32.24%		



difficult to discuss since arsenic was not significant in reducing growth but yet was significant in inhibiting per cent germination and rate of establishment as given on Table XIV. The high phosphorus level was again significant in promoting taller growth of all the grasses. Crabgrass grew significantly taller than any of the other grasses. Colonial bentgrass and red fescue were significantly taller than was either annual bluegrass or Kentucky bluegrass.

In summary, the overall averages for all measurements for the six-week seeding were less than for any of the previous seeding dates. This may have been due to climatic factors as well as to chemical treatments. Arsenic was toxic to both germination and rate of establishment but did not affect the growth in height. High phosphorus seemed to have little effect on germination and rate of establishment but was highly significant in promoting additional growth in height. The only big difference in grass response was that the red fescue appeared to be more tolerant to arsenate injury than in the previous seeding dates. Kentucky bluegrass was always the least aggressive but this is inherent to its nature in the seedling stage.

The original objective may be answered by stating: when calcium arsenate was applied as a pre-emergence herbicide on open soil, there was no significant interaction under different phosphorus levels. Grasses were different in their response to phosphorus and arsenate but did not respond consistently to either arsenate or phosphate when seeded at the time of treatment, three weeks and six weeks later.

TABLE XIV

AVERAGE HEIGHT IN CENTIMETERS OF GRASSES SEEDED  
SIX WEEKS AFTER ARSENATE TREATMENT  
UNDER TWO PHOSPHORUS LEVELS

Grasses	High Phosphorus		Low Phosphorus		Treatment Average		Species Average
	Arsenic Treated	Check	Arsenic Treated	Check	Arsenic Treated	Check	
Annual Bluegrass	1.2	2.2	1.5	2.0	1.4	2.1	1.8
Colonial Bentgrass	2.6	2.5	3.2	3.9	2.9	3.2	3.1
Red Fescue	2.9	3.2	3.9	3.9	3.4	3.6	3.5
Kentucky Bluegrass	2.1	2.0	3.5	2.0	2.8	2.0	2.4
Crabgrass	8.5	9.8	9.0	11.2	8.8	10.5	9.6
Average	<u>3.5</u>	<u>3.9</u>	<u>4.2</u>	<u>4.6</u>	<u>3.8</u>	<u>4.3</u>	4.1
P Level Av.	3.7		4.4				

## Summary of Analysis of Variance

Source of Variation	F Value	D Value 5%
Phosphorus Levels	12.1	0.6
Chemical Treatment	2.3	N.S.
Grass Species	102.5	1.2
Phosphorus x Chemical	0.1	N.S.
Phosphorus x Grass	0.4	N.S.
Chemical x Grass	2.2	N.S.
Phosphorus x Chemical x Grass	0.6	N.S.
Coefficient of Variability - 30.94%		

#### D. Chemical Analyses

1. Purpose. The purpose of the chemical analyses was to determine, in the five grasses, the assimilation of arsenic under two levels of phosphorus. Total phosphorus and total arsenic analyses were made for tops and roots of respective plots. Additional analyses of arsenic and phosphorus were made for the soil to ascertain whether there was downward movement of the arsenate under the two phosphorus levels.

2. Materials and Methods. In the summer of 1962, clippings were collected from the field plots treated and seeded on the same date. These were the first clippings taken from the plots. After drying, they were ground, first, in a Wiley Mill and, finally, prepared in a Micro Sampling Hammer Mill. The four separate replications of clippings were made into one composite sample and divided with a proportioner. The final proportion was used for analysis. In the fall of 1962, root samples from these plots were also taken. One sample from each plot was taken with a four-inch "green's cup set." A special screening board was used for washing and air drying the root samples. After removal of the crown, the roots were dried in a 105° oven. The samples were composited and ground in the Micro Sampling Mill.

The exact chemical methods employed will be discussed only briefly. The arsenic method used was the same for both tops and roots, however, a two gram sample of tops was used in contrast to one gram for the roots. The standard method for arsenic in plants was followed as given in Official Methods of Analysis by A.O.A.C. (14). The phosphorus method used was the same for both tops and roots and is given in the 1960 edition of A.O.A.C. Methods of Analysis (15).



Chemical analyses were made for arsenic and phosphorus in soils. Before the last seeding was carried out, one inch diameter cores were taken at zero- to one- and one- to two-inch depths. Representative samples were taken on the arsenic-treated and control plots under both levels of phosphorus. After air drying, they were ground in the Micro Sampling Hammer Mill. The chemical procedure used for arsenic is given in Official Methods of Analysis by A.O.A.C. (14). Total phosphorus was determined by the method given by Jackson (12). The exact procedure used in this case needs explanation. Jackson published two methods for total phosphorus. His method using perchloric acid was found to be best suited for this analysis. After digestion was completed, contents were made up to a 200 ml. volume. A 10 ml. aliquot was processed through a bromide distillation according to Jackson pg. 154 (12). This method was suggested where large amounts of arsenic are present. The determination was completed by using the vanadomolibdophosphoric color method for phosphorus. After color development, the yellow solution was measured on a Coleman spectrophotometer at 440 mu.

3. Results and Discussion. Chemical analyses many times help explain certain physiological responses of grasses. An effort was made in these chemical tests to observe two specific reactions. These were assimilation of arsenic, and assimilation of phosphorus by the five species of grasses.

A discussion of the root data will be presented first, as reported in Table XV. The total weight of dry matter for the grass roots was greater for the low than for the high phosphorus level but not significantly so. The quantity of arsenic found for the high or low phosphorus

TABLE XV

DRY WEIGHT AND CHEMICAL DETERMINATIONS OF THE CLIPPINGS AND  
ROOTS TAKEN FROM GRASS PLOTS SEEDED AND TREATED THE SAME  
DAY WITH ARSENATE UNDER TWO PHOSPHORUS LEVELS

Clippings of Grass Species	High Phosphorus					
	Calcium Arsenate			Check		
	Dry Wt. Grams	P ppm	As ppm	Dry Wt. Grams	P ppm	As ppm
Annual Bluegrass	266	6256	33	386	6242	20
Colonial Bentgrass	206	6252	33	250	6678	26
Red Fescue	198	6329	22	251	6853	14
Kentucky Bluegrass	172	6897	54	269	7595	14
Crabgrass	<u>318</u>	<u>6853</u>	<u>50</u>	<u>414</u>	<u>8294</u>	<u>29</u>
Average	232.0	6517.4	38.4	314.0	7132.4	20.6
<u>Roots of Grass Species</u>						
Annual Bluegrass	1.8	2969	74	1.3	2925	22
Colonial Bentgrass	3.2	2620	34	3.2	2489	8
Red Fescue	3.4	2576	44	4.6	2270	6
Kentucky Bluegrass	3.8	2620	33	4.0	2533	5
Crabgrass	<u>2.1</u>	<u>1877</u>	<u>20</u>	<u>2.1</u>	<u>1746</u>	<u>24</u>
Average	2.9	2532.4	41.0	3.0	2392.6	13.0

TABLE XV (continued)

Low Phosphorus						
Clippings of Grass Species	Calcium Arsenate			Check		
	Dry Wt. Grams	P ppm	As ppm	Dry Wt. Grams	P ppm	As ppm
Annual Bluegrass	138	4450	17	214	4843	21
Colonial Bentgrass	94	4952	35	152	4843	16
Red Fescue	94	4516	18	141	5214	28
Kentucky Bluegrass	87	5558	17	127	5126	24
Crabgrass	<u>127</u>	<u>3973</u>	<u>69</u>	<u>245</u>	<u>3755</u>	<u>20</u>
Average	108.0	4689.8	31.2	175.8	4774.0	21.8
<u>Roots of Grass Species</u>						
Annual Bluegrass	4.1	2489	134	2.4	2620	12
Colonial Bentgrass	2.5	2533	58	1.7	2445	16
Red Fescue	4.6	2358	30	3.5	2270	7
Kentucky Bluegrass	2.8	2533	31	4.4	2183	9
Crabgrass	<u>6.3</u>	<u>1746</u>	<u>83</u>	<u>2.7</u>	<u>2358</u>	<u>6</u>
Average	4.1	2331.8	67.2	2.9	2375.2	10.0
Summary of Averages						
	Clippings			Dry Wt.	Roots	
	Dry Wt.	ppm P	ppm As		ppm P	ppm As
High P	273.0	6824.1	29.5	2.9	2462.5	27.0
Low P	141.9	4723.0	26.4	3.5	2353.5	38.6
T Value 5%	4.4	7.6	N.S.	N.S.	N.S.	N.S.



levels was also statistically non-significant as was the amount of phosphorus. In discussing the clipping analyses, the only significant difference was that grass grown in high phosphorus soils yielded more dry matter than those under low phosphorus. When these clippings were analyzed, those from the high phosphorus soils were significantly greater in phosphorus content than were the low phosphorus clippings. Arsenic assimilation was not significant under either phosphorus level.

Much of the literature reviewed indicated that arsenic assimilation by plants would be directly related to phosphorus content in the soil. Four simple correlations were computed and found to have the following coefficients: 1)  $-.288$  for arsenic x phosphorus in clippings under low phosphorus soils, 2)  $-.149$  for the same factors in roots grown in low phosphorus soil, 3)  $-.106$  for arsenic x phosphorus in tops under high phosphorus soils, and finally, 4)  $.189$  for arsenic x phosphorus in roots under high phosphorus. As may be observed in Table XV, the ratio of arsenic to phosphorus is not close to the toxic point for plants according to Hurd-Karrer (11). She states that a 1:1 ratio was required before death would occur. This may not be a true comparison, however, since her ratio was derived after intensive study with nutrient cultures. Everett (8) found that plants in solution cultures react quite differently than in soils. The point made is that in these tests under soil conditions, neither phosphorus level was significant in causing a response in the arsenic assimilation. The average for arsenic content in the roots under low phosphorus was found to be higher but not significantly so. By looking at the results of the soil test, the former discussion may be explained.

Chemical analyses were made for the field plot soils to determine

the quantities of arsenic and phosphorus in the upper zero-to one- and one-to two-inch soil levels. Eight different soils were analyzed and the results are given in Table XVI. As reported by Swenson, Cole and Sieling (22), when high phosphorus levels are present in a soil over an extended period of time, the chances of also finding a high arsenate level is highly improbable since anion exchange would displace the arsenic ion to lower horizons, however, this was not the trend found in these soils. A possible reason for this is that, since phosphorus and arsenic react and move very slowly in soil, the time lapse between arsenic application and time of testing was insufficient for the arsenic to become soluble. This may be noted in the ppm of arsenic still remaining in the upper one inch of soil. This may also be a good explanation why more injury due to arsenic toxicity does not occur on the established turf grasses.

In summary, for the chemical tests it may be stated that no trend of interaction occurred in respect to phosphorus inhibiting the uptake of arsenic or the movement of arsenic in soil. It is believed that arsenic was not in sufficiently available form to have any great effect on the five grasses grown on the treated soils.

TABLE XVI

CHEMICAL DETERMINATION FOR PHOSPHORUS AND ARSENIC IN SOILS  
TAKEN FROM ARSENIC TREATED AND CONTROL PLOTS  
SIX WEEKS AFTER APPLICATION

Soil Sample	Arsenic Plots		Control Plots	
	As ppm	P ppm	As ppm	P ppm
High P, 2" depth	76.0	1620	24.5	1600
High P, 1" depth	480.0	1640	24.0	1261
Low P, 2" depth	76.0	1120	29.0	1740
Low P, 1" depth	395.0	1100	20.0	1200



#### IV. CONCLUSIONS

##### Greenhouse Test

1) Copper arsenate was not as effective in controlling annual bluegrass and crabgrass as was calcium arsenate under the various levels of phosphorus studied.

2) Calcium arsenate reduced plant growth as well at 391 pounds per acre as it did at 793 pounds per acre.

3) The silt loam and sandy loam soils were equally good in supporting plant growth under all arsenic treatments and phosphorus levels.

4) No detectable significant interaction occurred between arsenate and phosphorus as measured by grass response.

##### Field Test - (a) Seeded and treated same day.

1) The action of arsenic was independent of either phosphorus level.

2) The high phosphorus soil level was significantly better in promoting establishment and growth in height than the soil of low phosphorus content.

3) Arsenic significantly reduced germination, rate of establishment and growth in height. Since 100 per cent control of annual bluegrass and crabgrass is desired, the above reduction is of little practical consequence. Since injury to the three seedling turf grasses occurred,

reservation is held as to the practicality of arsenate treatment under either high or low phosphorus soil levels in seedling establishment.

(b) Seeded three weeks after arsenate application.

1) There were no consistent trends in this test with respect to arsenic toxicity.

2) The high phosphorus soil level was significant in promoting good establishment and growth in height of grass species over the low phosphorus level.

3) No interactions of phosphorus levels and arsenate treatment were observed.

(c) Seeded six weeks after arsenic application.

1) A reduction in overall average was observed when compared with overall average of tests (a) and (b).

2) The high phosphorus soil level was significant in promoting growth in height.

3) No particular trend was prevalent for grasses in any of the measurements except where crabgrass and, at times, annual bluegrass, were inhibited more by the arsenic treatment than were the basic turf grasses.

#### Chemical Determinations

1) The phosphorus, as measured, did not significantly influence the assimilation of arsenic into the grass plants.

2) The high phosphorus soil level was significant in promoting a greater dry matter and phosphorus content than was the low phosphorus level.

3) Neither high nor low phosphorus influenced differential movement of arsenic into lower horizons. Approximately 20 per cent of the applied arsenate remained in the two-inch level.

A study was started in 1941 to determine whether various levels of soil phosphorus would affect the herbicidal properties of calcium and copper arsenates. The investigation was divided into three separate parts. The first was a greenhouse study, the second a field test, and the third consisted of chemical analysis of plants and soils from the field study.

The greenhouse tests were set up by using four-inch plastic pots arranged in blocks with annual blunders (*Portulaca L.*) and morning glories (*Ipomoea pes-caprae* Benth.) planted on two different soil types. The soil types were separated into three partitions representing low, medium and high levels of phosphorus, respectively. Copper arsenate and calcium arsenate were used at high and low rates on each of the three phosphorus levels. It was found that the low rate of calcium arsenate at 0.5 lb. per square yard, was as effective in controlling morning glories and annual blunders as were the other treatments. Soil type and phosphorus levels did not have a marked effect on control injury.

In the summer of 1941, a field test was conducted on the University of Idaho Forest Experiment Station near Moscow, Idaho. The purpose of this test was to determine the response of five species to a single conventional rate of calcium arsenate under the levels of phosphorus. The ground was divided into three parts representing low, medium and high levels of phosphorus. The plants used were *Portulaca L.*, *Ipomoea pes-caprae* Benth., *Portulaca L.*, *Ipomoea pes-caprae* Benth., *Portulaca L.*, and *Ipomoea pes-caprae* Benth.



## V. SUMMARY

A study was started in 1961 to determine whether various levels of soil phosphorus would affect the herbicidal properties of calcium and copper arsenates. The investigation was divided into three separate tests. The first was a greenhouse study, the second a field test, and the third consisted of chemical analyses of plants and soils from the field study.

The greenhouse test was set up by using four-inch plastic pots arranged in blocks with annual bluegrass (Poa annua L.) and crabgrass (Digitaria ischaemum Schreb.) planted on two different soil types. The soil types were separated into three partitions representing low, medium and high levels of phosphorus, respectively. Copper arsenate and calcium arsenate were used at high and low rates on each of the three phosphorus levels. It was found that the low rate of calcium arsenate at 391 pounds per acre, was as effective in controlling crabgrass and annual bluegrass as were the other treatments. Soil types and phosphorus levels did not have a marked effect on arsenic injury.

In the summer of 1962, a field test was established on the University of Rhode Island Agricultural Experiment Station Turfgrass Plots. The purpose of this test was to determine the response of five grasses to a single conventional rate of calcium arsenate under two levels of phosphorus. The grasses used were annual bluegrass (Poa annua L.), colonial bentgrass (Agrostis tenuis Sibth.), red fescue (Festuca rubra L.), Kentucky bluegrass (Poa pratensis L.), and smooth

crabgrass (Digitaria ischaemum Schreb.). Each grass was (a) seeded and treated with arsenic the same day, (b) seeded three weeks after arsenic treatment, and finally, (c) seeded six weeks after the arsenate application. For each seeding date three types of readings were observed, namely; germination, rate of establishment recorded as per cent cover and height of grass growth. The results indicate that phosphorus level had no detectable effect in arsenic inhibition. High phosphorus in general promoted better cover and growth over low phosphorus but had little effect on germination. Grasses differed significantly in response to phosphorus levels and arsenic treatment. Crabgrass and annual bluegrass showed the greatest growth under the high phosphorus levels and the least inhibition to the arsenic treatment. Calcium arsenate reduced germination, rate of establishment and height in growth significantly; however, the results are not practical for use in field maintenance.

The chemical tests consisted of making arsenic and phosphorus determinations in roots and tops from plots (a) seeded and treated with arsenic the same day. The purpose of these tests was to observe the effect of phosphorus levels on arsenic activity in respect to assimilation by the grass plants. Soil tests for total arsenic and total phosphorus were also made to observe whether phosphorus had any differential influence on the movement of arsenic in the soil. It was found that phosphorus did not affect the assimilation of arsenate. The soil test gave added evidence that neither high nor low phosphorus levels affected the movement of arsenic downward. Specific reasons for such a reaction were not analyzed because of the vast number of soil reactions which could be responsible for such phenomenon as stated in the literature, but the short period covered by the experiment may have allowed too little time for effective reactions.

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